

GW26-e1038**MicroRNA-20a/b Regulates Cholesterol Efflux through Posttranscriptional Repression of ATP-Binding Cassette Transporter A1 in THP-1 macrophage/RAW 264.7-derived foam cells**

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OBJECTIVES ATP-binding cassette transporter A1 (ABCA1) plays a crucial role in reverse cholesterol transport and has anti-atherosclerosis effects. It has been reported that microRNAs regulate the expression of ABCA1. Recent studies have shown that microRNA (miR)-20a/b might play an important role in atherosclerotic disease. A combination of bioinformatic tools for miRNA target prediction revealed that miR-20 family that is highly conserved directly targets the ABCA1 3' UTR. In this study, we attempted to clarify the effect of miR-20a/b on expression of ABCA1 and cholesterol efflux.

METHODS Luciferase reporter assays were used to confirm whether ABCA1 was a target of miR-20a/b. ABCA1 expression was measured by real-time quantitative PCR and western blot analyses. Cellular cholesterol efflux was analyzed using liquid scintillation counting assays. High Performance Liquid Chromatography was performed to determine the cellular total cholesterol and free cholesterol.

RESULTS MiR-20a/b significantly reduced the luciferase activity of the ABCA1 3' UTR reporter construct ($P < 0.05$). MiR-20a/b decreased the ABCA1 expression in THP-1 macrophage/RAW 264.7-derived foam cells, which in turn reduced apoA-I mediated cholesterol efflux by 30% to 40% and increase cholesterol content ($P < 0.05$). On the contrary, miR-20a/b inhibitor can increase the ABCA1 expression, increase the cholesterol efflux by 30% to 35%, decrease cholesterol content ($P < 0.05$), inhibit foam cell formation and protect against atherosclerosis.

CONCLUSIONS MiR-20 is a new miRNA that can target ABCA1, regulate ABCA1 expression and regulate atherosclerosis.

GW26-e0808**Sodium tanshinone IIA sulfonate inhibits angiotensin II induced extracellular matrix remodelling in cardiac fibroblasts -Implications for treatment of pathologic cardiac remodelling**

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OBJECTIVES Cardiac extracellular matrix (ECM) remodeling involves fibroblasts differentiation, elevated matrix and metalloproteinase (MMPs) production and is critical in the pathogenesis of cardiac hypertension and heart failure. Cytokines such as angiotensin II, play a pivotal role in these pathogenic processes. Sodium tanshinone IIA sulfonate has been reported to alleviate the maladaptive effects of left ventricular hypertrophy developed in animals models of myocardial infarction and suggested that this effect could be partially linked to the increased expression of matrix metalloproteinase-1. However, the mechanism by which sodium tanshinone IIA sulfonate exerts these beneficial effects, specifically how it modulates the production of ECM and MMPs has yet to be fully characterized.

METHODS Our present studies, performed on cultures of cardiac fibroblasts demonstrated that angiotensin II induced a significant increase in the expression of collagen I, collagen III and fibronectin.

RESULTS This effect was inhibited by pretreatment with sodium tanshinone IIA sulfonate. The underlying mechanism rests in sodium tanshinone IIA sulfonate's ability to suppress cardiac fibroblast differentiation and inhibitions of the angiotensin II-induced Smad pathway. Specifically, sodium tanshinone IIA sulfonate treatment reduced the binding capacity of CREB-binding protein 1 (CBP1), a transcriptional co-activator, to the phospho-Smad2-Smad4 transcription complex. Furthermore, sodium tanshinone IIA sulfonate attenuated angiotensin II-induced MMP9 expression following inhibition of ERK1/2 phosphorylation and NF- κ B p65 nuclear translocation.

CONCLUSIONS Together, these findings implicate sodium tanshinone IIA sulfonate as a critical mediator in suppressing cardiac matrix remodeling, principally through its inhibitory effects on Smad signaling and recruitment of coactivators to Smad transcriptional

complexes. Concurrently, sodium tanshinone IIA sulfonate reduced the expression of MMP9, which is associated with the inhibition of ERK and NF- κ B pathway.

GW26-e1074**High number of transplanted stem cells improves myocardial recovery after acute myocardial infarction in rats**

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OBJECTIVES Our objective was to study the cardiac function and changes in cytokine levels after administration of BMMNC in experimental acute myocardial infarction (AMI) model.

METHODS Unlabeled or Super-Paramagnetic-Iron-Oxide-labeled bone marrow mononuclear cell (BMMNC) or saline was injected into myocardium of 30 rats after AMI. Ejection fraction (EF) was measured preoperatively, postoperatively and at 21 days by echocardiography. Cardiac MRI was performed postoperatively and after 21 days in 7 BMMNC animals. Serum cytokine levels were measured at baseline, 24 h and 21 days. Cellular homing was evaluated comparing MRI and histology.

RESULTS From baseline to 21 days EF decreased less in BMMNC group (EF mean control -19 SD 12 vs. BMMNC -4 SD 15 percentage points $p = 0.02$). Cytokine concentrations showed high variability between the animals. MRI correlated with histology in cell detection and revealed BMMNCs in the infarction area. By MRI, EF improved 12 percentage points. The improvement in EF was associated with the number of transplanted BMMNCs detected in the myocardium.

CONCLUSIONS BMMNC injection after AMI improved cardiac function. Quantity of transplanted BMMNCs correlated with the improvement in cardiac function after AMI.

GW26-e4395**Collaboration of HIF-2 α and Oct4 Promotes Survival and Myocardia Repair of Human Very Small Embryonic-like Mesenchymal Stem Cells In Myocardial Infarction**

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OBJECTIVES The effects of mesenchymal stem cells therapy on myocardial infarct (MI) might be affected by their low survival and functions, which were regulated by the family of hypoxia inducible factors (HIFs). The transcription factor HIF-2 α is an essential regulator of transcriptional response to hypoxia, can interact with embryonic stem cells (ESCs) transcription factor Oct4 and modulate its signaling.

METHODS Here, we obtained a very small embryonic-like mesenchymal stem cells (vseMSCs) from the MI patients, which possessed the VSELs' morphology as well as ESCs' pluripotency.

RESULTS Using microarray analysis, we compared HIF-2 α -regulated gene profiles in vseMSCs to ES cell profiles and determined that HIF-2 α coexpressed Oct4 in vseMSCs similarly to ES cells. However, this coexpression was absent in unpurified MSCs (uMSCs). Under hypoxic condition, vseMSCs exhibited stronger survival, proliferation, and differentiation than uMSCs. Transplantation of vseMSCs caused the greatest improvement of cardiac function and heart remodeling in the infarcted rats. We further demonstrated that HIF-2 α and Oct4 jointly regulate their relative downstream gene expression, including Bcl2 and Survivin, which were essential for vseMSCs survival and proliferation, Nanog, Klf4, and Sox2, which are important pluripotent markers, and Ang-1, bFGF, and VEGF, which promoted angiogenesis. Importantly, these effects were generally magnified by up-regulation of HIF-2 α and Oct4 induced by HIF-2 α or Oct4 overexpression and abolished by HIF-2 α or Oct4 deficiency.

CONCLUSIONS Together, these findings demonstrate that HIF-2 α in vseMSCs interacts with Oct4 in survival and function. The identification of the actions between HIF-2 α and Oct4 will enable deeper characterization of the downstream targets of this interaction in vseMSCs and novel pathophysiological implications for repair of infarcted myocardium.